IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s): Maurer, et al.	
Application No.: 10/019,199	
Filed: 12/20/2001	Group Art Unit: 1615
Title: Methods for Preparation of Lipid-Encapsulated Therapeutic Agents	Examiner: G.S. Kishore
Attorney Docket No.: INEX.P-005	Confirmation No: 6234
Customer No.: 021121	

Commissioner for Patents
PO Box 1450
Alexandria, VA 22313-1450

RESPONSE TO OFFICIAL ACTION

Dear Sir:

This is in response to the Office Action mailed December 30, 2003 for the above-captioned application. Reconsideration and further examination are respectfully requested.

Applicants request an extension of time sufficient to make this paper timely and enclose the fee. The Commissioner is authorized to charge any additional fees or credit any overpayment to Deposit Account No. 15-0610.

Claims 13-32 are pending in this application.

I hereby certify that this paper and any attachments named herein are transmitted to the United States Patent and Trademark Office, Fax number: 703-872-9306 on April 30, 2004.

Marina T Larson

Marina T. Larson, PTO Reg. No. 32,038

April 30, 2004
Date of Signature

Appln No.: 10/019,199
Amendment Dated: April 30, 2004
Reply to Office Action of December 30, 2003

The Examiner finally rejected claims 13-32 under 35 USC § 103 as obvious over the combination of US Patent No. 6,447,800 of Hope et al. and US Patent No. 5,976,567 of Wheeler or WO 98/51278. Applicants respectfully traverse this rejection, and enclose a Declaration Under Rule 132 addressing the differences between the Hope and Wheeler references, and the presently claimed invention. This declaration is executed by Dr. Michael Hope, who is a named inventor on each of the references cited as well as a person of skill in the relevant art. (See Declaration, ¶¶ 2-5).

As observed in the declaration (¶ 7), and as reflected in the claims, the claims of this application are directed to a method for preparing lipid particles that contain a fully-encapsulated charged therapeutic agent such as an oligonucleotide or polynucleotide. The product formed in this method therefore has the charged therapeutic agent on the inside of the lipid particles, encapsulated in and protected by the lipid. The method involves taking pre-formed lipid vesicles, and combining them with the charged therapeutic agent and a destabilizing agent and incubating for a period of time sufficient to allow encapsulation of the therapeutic agent. Thereafter, the destabilizing agent is removed. The preformed lipid vesicles comprise at least two lipid components: a charged lipid that is opposite in charge to the therapeutic agent, and a modified lipid having steric barrier properties.

The Examiner asserts that Hope teaches permeabilization of a lipid membrane with ethanol to allow encapsulation, but acknowledges that Hope does not disclose the use of a charged lipid or removal of ethanol. Wheeler is cited to address this failure because in this patent, ethanol is removed from lipid preparation that include cationic lipids. The legal question is whether this relationship is real, such that a person skilled in the art would consider making the combination of elements proposed by the Examiner, or merely a superficial similarity that offers no suggestion of such a combination.

As stated in the declaration (¶ 8), Dr. Hope, as both an author of the cited references and a person skilled in the art, "believe that the Examiner is taking selected parts from each of these references in a manner which would not be apparent absent the guidance of the present application." The reasons for this are set out in the declaration.

The Hope patent teaches a method for loading materials, including therapeutic agents, into liposomes by rendering a preformed liposomal membrane permeable using an organic solvent, preferably ethanol. (Declaration, ¶ 9) The key to the Hope patent is that ethanol permeabilizes the membrane without changing the structure. The ethanol establishes an organic-solvent induced permeation gradient (col 8, lines 34-65). In order to effectively trap the material in the liposome, after membrane permeation, the permeability barrier must be restored. This is generally accomplished by diluting the solvent (col 9, line 22).

The therapeutic agents used in the method of the present invention are charged. The Hope patent states that "generally highly negatively charged species such as polynucleotides do not cross the liposomal membranes permeabilized by the solvent technique disclosed herein." (Col. 10, lines 7-9). (Declaration, ¶ 12). Indeed, based on experiments conducted in 1990-1992 by Dr. Hope, the technique disclosed in the Hope patent does not work with charged oligonucleotides. (Declaration, ¶ 11).

In contrast to the operation of the Hope patent, the nucleic acid loading method disclosed in Wheeler restructures the lipid material by dissolving it in a solvent. (Declaration, ¶ 9). US Patent No. 5,976,567 of Wheeler relates to the formation of lipid-nucleic acid complexes. The method makes use of ethanol, but this is not a point of similarity with the Hope patent, because the ethanol is not used to permeabilize a pre-formed lipid membrane. There is no disclosure in Wheeler of introducing nucleic acids, or any other charged material through the membrane of a pre-formed lipid particle. Instead, as reflected in Fig. 40 of Wheeler, formation of the particles results from rearrangement of the lipid membrane or from a coating of particles onto the DNA. (Declaration, ¶ 13).

Furthermore, Applicants again note that we know from the Hope patent that negatively-charged species do not pass through an uncharged membrane. Why then would a person skilled in the art imagine that negatively charged polynucleotides would pass through a membrane better, when that membrane contains positive charges to which they can stick as shown in Wheeler? Applicants submit that, "without knowledge of the present invention, the idea of adding charged lipids of opposite charge to the therapeutic agent would not be a reasonable option, since one skilled in the art would reasonably expect that adding positive charges in the lipid would cause negatively charged lipid to bind to the lipid, making passage through the membrane more, not less, difficult." (Declaration ¶ 12). Thus, this is a further reason why the combination of references advanced by the Examiner would not be suggested by the references to a person skilled in the art.

From these explanations, it can be seen that the basis for the combination of the Hope and Wheeler reference is really based only on the superficial similarity in words and materials, and not on a real similarity which would provide motivation to a person skilled in the art. The types of lipid structures and the physical changes that the lipid structures undergo in the two applications are different. One involves transport of materials through an uncharged membrane, while the other involves formation of complex based on interactions of charged species. (Declaration, ¶¶ 9 and 16) Further, the role of ethanol in the two references is different. (Declaration, ¶ 13). The purposes of the lipid structures in the Hope and the Wehhler references are different. (Declaration, ¶ 15) Thus, it can be seen that the rejection is based solely on finding the isolated elements, and that the motivation to make the combination is lacking. This is not enough, since "citing references which merely indicate the isolated elements ... are known is not

a sufficient basis for concluding that the combination of elements would have been obvious." *Ex Parte Hiyamizu*, 10 USPQ 2d 1393, 1394 (POBAI 1988)

With respect to the third reference, Applicants note that the Examiner has said that the addition of PEG-lipids to the compositions of Hope would have been obvious since WO 98/51278 teaches their ability to provide steric stabilization. What the Examiner has not said, is why one skilled in the art would think that "steric stablization" is needed in the liposomes of Hope. Steric stabilization is used in the reference to reduce or eliminate aggregation of lipid particles. Nothing in Hope indicates that such aggregation would be a problem **in the context of the Hope reference**, nor has the Examiner offered any reasoning as to why a person skilled in the art would anticipate the existence of such problems **in compositions such as those of Hope**. The Examiner must find motivation in the context of the art, not in the abstract, in order to avoid the unreasoned selection of elements. Here that has not been done. Accordingly, there is no motivation in the cited art to include PEG-lipids in the compositions of Hope. Applicants note that the Examiner asserts that "aggregation will be a problem whether recognized by Hope or not," but has not said why this would be the case in uncharged liposomes with uncharged materials encapsulated therein. There is no evidence of record that aggregation is a problem known to be an issue for all liposomes lacking a PEG lipid, regardless of their chemical makeup.

Applicants also point out once again that the claims call for an intermediate of modified lipid that prevents aggregation without completely eliminating it. This intermediate amount is important to the success of the method of the present invention but is not suggested in the references. Nor is there any indication in the cited references that this would be a result-affecting parameter. As noted in the application (Page 13 and Example 7), the formation of the particles of the invention appears to involve an aggregation step, i.e. it is a structural organization process and not a simple permeability effect as in HOPE, and is not a process of simple passage of the charged material, such as a polynucleotide, through the membrane. (Declaration, ¶ 17).

Finally, Applicants point out that the method of the invention provide advantages that cannot be predicted based on the art. The ability to introduce charged therapeutic agents into a pre-formed lipid particle, without rearrangement is important, because the size of the resulting particle is predictable because it in essence remains unchanged. The size of a particle containing a therapeutic agent can be a significant factor in toxicity and clearance rate of the particles. Thus, the ability to control it predictably is important. This control is lacking in Wheeler, as Wheeler restructures the lipid material (which is not a liposome). In a manufacturing process using the current method, one can add material to the interior of the liposomes without changing the liposome. (Declaration, ¶ 14).

The Examiner also rejected claims 13-32 as unpatentable over Hope in view of Malone and Zalipsky. This rejection is very similar to that discussed above, with Malone providing the

teaching of cationic lipid nucleic acid complexes, and Zalipsky offering a teaching concerning PEG-lipids. This rejection is deficient for the same reasons as discussed above.

The Examiner also rejected claims13-20 and 25-32 as unpatentable over Schubert in view of Malone and either Zalipsky or WO 98/51278. Schubert is substantially cumulative with Hope, except that a different method for opening the membrane of a pre-formed liposome is disclosed. Specifically, in the case of Schubert, sodium cholate, a bile salt, is used to open the membrane. The Examiner has not indicated why a person skilled in the art would anticipate that changing the liposome structure to include a cationic lipid would allow loading of pre-formed lipids with a negatively charged oligonucleotide, without this step of membrane opening being necessary. In this regard, it is noted that the cholate part of the bile salt has a negative charge. If one introduced a cationic lipid into the liposome of Schubert, there is no basis provided in the reference or the Examiner's argument, to conclude that the cholate would still function to open the membrane. To the contrary, it might well associate with the positive charges of the cationic lipid, yielding a wholly different result. Thus, it is naive and overly simplistic to argue, as the Examiner has done, that cholate makes holes, and cationic lipids allow transfection, and that we can put the two features together in a single species without concern or thought for the interactions of the cholate and the cationic lipid. Further, the reliance on Malone or Zalipsky in this rejection is flawed for the same reasons as discussed above.

For these reasons, Applicants submit that the rejections of record should be withdrawn. This application is now considered to be in condition for allowance and such action is earnestly solicited.

Respectfully Submitted,

Marina T. Larson, Ph.D.
Attorney/Agent for Applicant(s)
Reg. No. 32038

(970) 468 6600

Attachments:
Rule 132 Declaration of Michael J. Hope
Request for Extension of Time
Credit Card Form